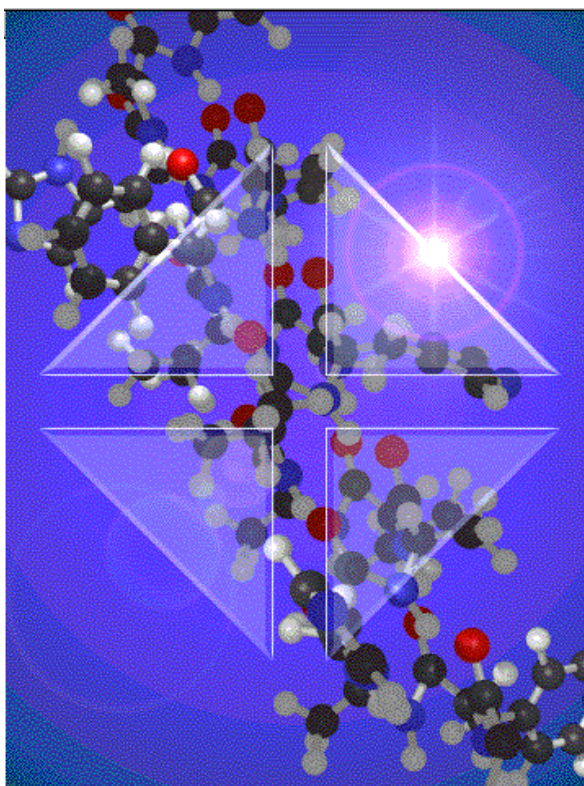


# UNICORN Version 3.20

Frac-950 module for UNICORN applications



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# 1 Frac-950 Setup and Documentation

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## 1.1 Introduction

UNICORN™ is a control system developed and marketed by Amersham Biosciences AB for real-time control of chromatographic separation systems from a personal computer. The package operates together with systems in the ÄKTA™*design* platform (referred to as ÄKTA*design* systems), BioPilot™ System and BioProcess™ System from Amersham Biosciences. Standard configurations are provided for ÄKTA*design* systems and BioPilot System, while configurations for process-scale separation with BioProcess System may be customised for a particular installation. UNICORN runs under the operating system Microsoft™ Windows NT™ version 4.0.

This manual describes the functions of the fraction collector Frac-950 within UNICORN 3.20. For a detailed background on other UNICORN functions, the user is referred to the UNICORN 3.10 manual (for a detailed description of the Frac-950 functions, see the Fraction collector Frac-950 user manual).

Functional features of Frac-950 include:

- User-defined rack choice, including tubes and microtubes.
- A variety of sampling orders during the fractionation process.
- Method templates, providing method frameworks for fractionation applications, eliminating the need to program methods from scratch.

For the purposes of this user manual, examples are displayed for an ÄKTA*explorer* 100 system. You should refer to the user instructions specific to the chromatography system that you are using.

In this manual, menu commands, the names of dialogue boxes and windows, the contents of dialogue boxes windows, and option buttons are written with a **bold helvetica** typeface. Menu commands are written in the order of the menu name and then the command, separated by a colon. For example:

“Select **File:Save As** to display the **Save As** dialogue. Locate the destination drive and folder and enter a file name. Click on **Save**.”

This directs you to click on the **File** menu and select the command, **Save As**. A dialogue called **Save As** is displayed in which you must locate the destination folder for the saved file and give the file a name. You then click on the button called **Save** in the dialogue to execute the save command.

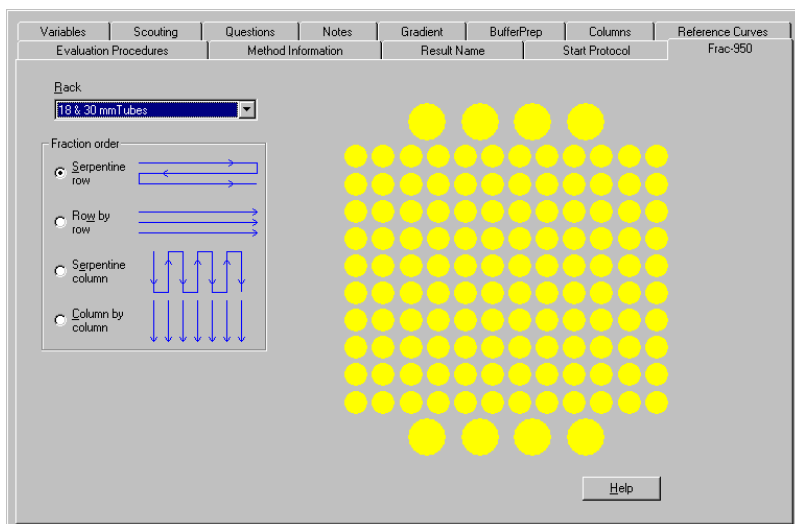
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## 1.2 Method editor: Run Setup



User-defined options for Frac-950 are selected on the Frac-950 tab in Run Setup in the Method editor. To reach Method editor from the Main menu, create a new method as described in the UNICORN 3.10 manual. To access the **Frac-950** tab in Run setup, either click on the Run setup icon on the toolbar, or select **View:Run Setup** from the menu. The **Frac-950** tab allows the user to choose rack type, the fractionation order, and the last tube position

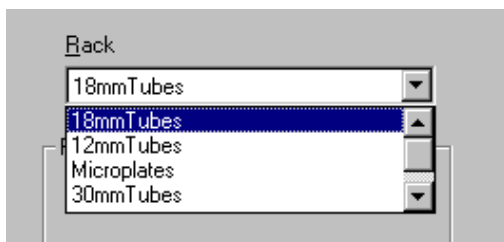
**Note:** The Frac-950 tab is only available if your strategy supports Frac-950.



**Figure 1-1.** The Frac-950 tab.

### 1.2.1 Rack type

Begin the fractionation set up by choosing the appropriate rack type from the **Rack** drop-down menu.

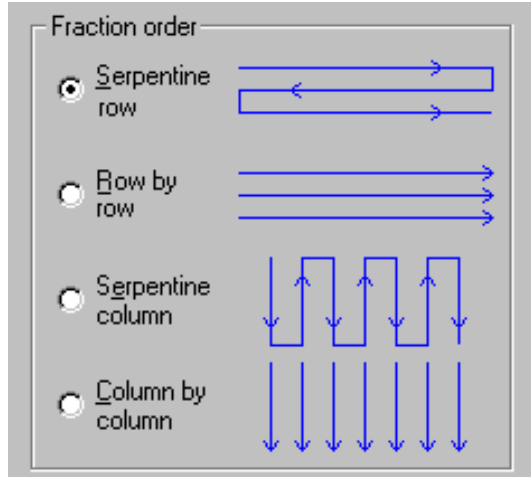


**Figure 1-2.** The Rack drop down menu within the Frac-950 tab.

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## 1.2.2 Fraction order

Next, use the **Fraction order** radio buttons to choose the appropriate order for fractionation.



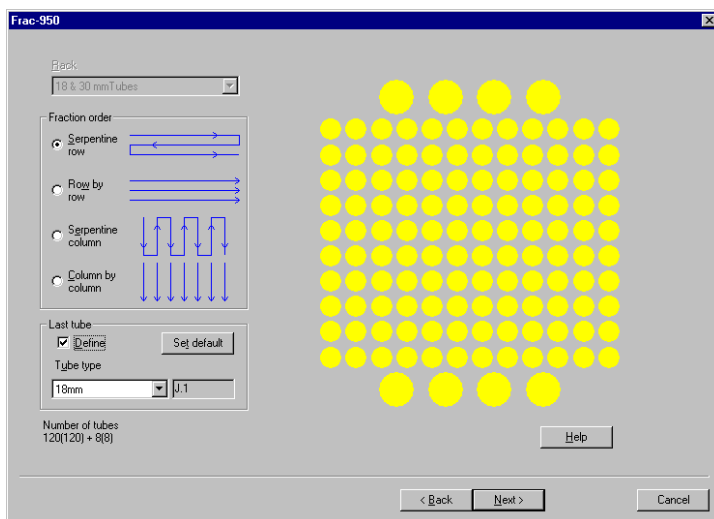
**Figure 1-3.** The Fraction order radio buttons.

## 1.3 System control

In System control, for manual runs, the **Frac-950** tab cannot be used in Documentation. Instead, use the manual fractionation instructions, starting with Man\_ (see also Appendix A, Manual control).

### 1.3.1 Last tube

The last tube position can only be selected from within **Start Protocol** before starting a run. Click the **Define** box within the **Last tube** panel, and then move the cursor over the appropriate tube (circle) within the tube matrix and click again.



**Figure 1-4.** The Frac-950 tab, illustrating the Last tube function.

If the fractionation process proceeds beyond this position during a run, an alarm will be executed. The lower right box within the **Last tube** panel shows the currently selected last tube.

If a new last tube has been chosen, but the user wishes to return to the default last tube position, simply click on the **Set default** button. This will automatically set the last tube to its default position.

**Note:** When using different sized tubes (racks: 18mm tubes or Microplates) the last tube can be set for both tube sizes. Use the **Tube type** drop down menu to choose the desired tube size, and then follow the procedures outline above to select the last tube.

Note that the total number of tubes sampled may differ according to which **Fraction order** has been chosen. The **Number of tubes** equation in the bottom left corner of the **Frac-950** tab shows the current number of available tubes chosen for fractionation, followed by the total possible number in parentheses. For example, in the figure above, 120 of the total 120 small tubes, and all 8 of the large tubes will be available for fractionation.

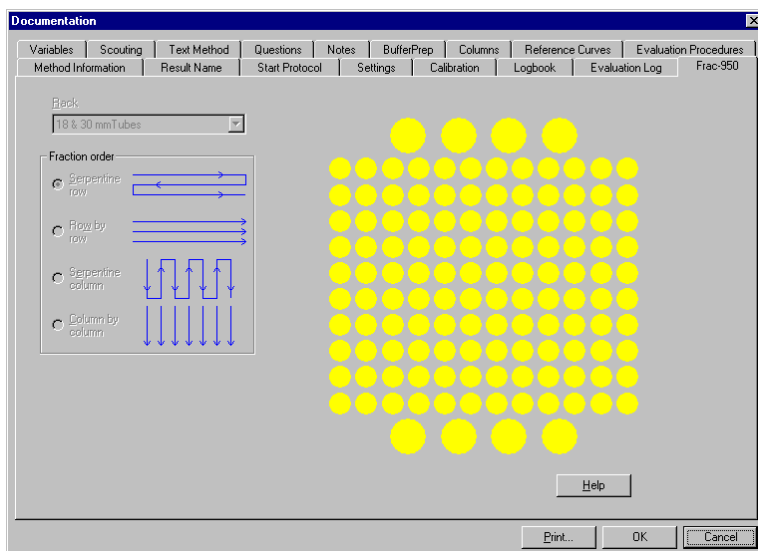
## 1.4 Evaluation

### 1.4.1 Documentation



The Frac-950 tab can be displayed within the Evaluation module by

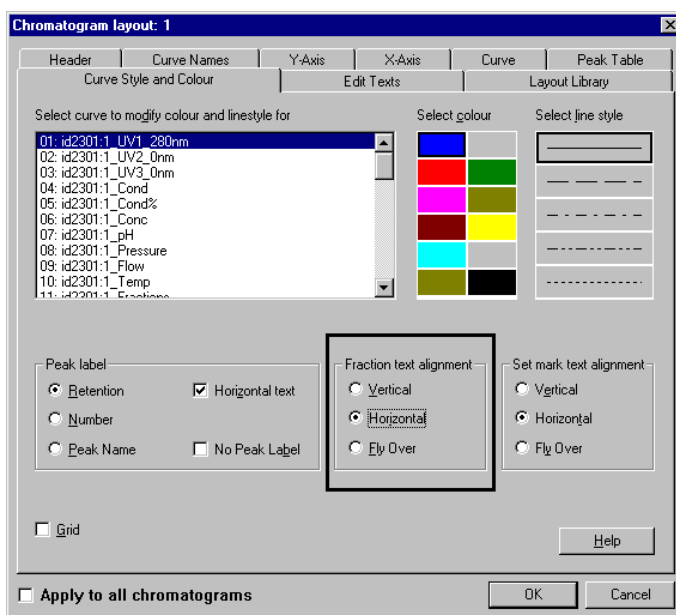
choosing **View:Documentation**, or pressing the Documentation icon on the toolbar, and pressing the **Frac-950** tab. You can print out the selected settings by pressing the **Print** button, or return to the main Evaluation window by pressing **OK** or **Cancel**.



**Figure 1-5.** The Frac-950 tab in Documentation in the Evaluation module.

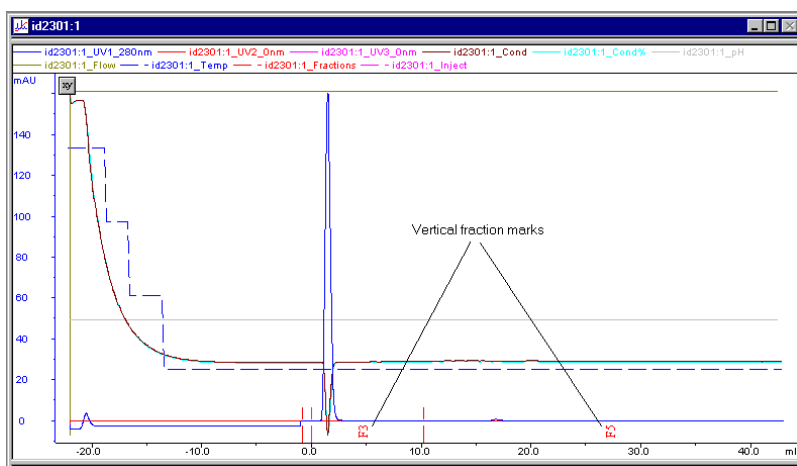
## 1.4.2 Fraction text alignment

In the **Evaluation** module, click on the chromatogram with the right-hand mouse button, select **Properties...** from the menu and then select the **Curve Style and Colour** tab. This will allow you to select **Vertical**, **Horizontal**, or **Fly Over** orientation for the Fraction text mark.



**Figure 1-6.** The chromatogram layout dialogue, with the Fraction text alignment box highlighted.

To change the fraction text alignment from the default option (horizontal), select the appropriate radio button, and then click on the **OK** button.



**Figure 1-7.** A chromatogram window in the Evaluation module, showing Vertical fraction text mark alignment.

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## A Strategy for ÄKTAexplorer 100

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A dedicated strategy is available for each system in the ÄKTA*design* platform. The instructions available differ slightly between strategies, although the majority are common. To find a description of any instruction in your particular strategy, mark the instruction and click on <F1>. To obtain a printout of the instructions, select **File:Print** in the Method editor and click on the **Instruction set** option box.

As an example, this appendix lists the instructions for methods, manual control and system settings supported by the strategy for ÄKTA*explorer* 100 with Frac-950 (E100F200).

### A.1 Method instructions

#### A.1.1 Pump

<i>Instruction</i>	<i>Description</i>	<i>Parameters</i>
Flow	Starts the flow for the gradient pump at the set flow rate	Flow rate (0-100 ml/min)
Gradient	Defines a linear gradient for gradient pump (pump A and B). Reaches the target value after the specified length. Length is expressed in time, volume or column volume depending on selected base.	Target (0-100 %B) Length (0-9999 base)
BufferPrep_pH	Selects pH for a defined BufferPrep recipe. When BufferPrep is not used, the instruction should be set to zero, otherwise an alarm will be given.	pH (0-14)
SampleFlow	Starts the flow of the sample pump at the set flow rate. The sample pump can be calibrated in the calibration module.	Flow rate (0-5 ml/min)

DirectLoad	The sample is directly loaded onto the column. ReduceFlow makes it possible to automatically reduce the flow during sample loading to avoid a pressure alarm. An alarm will be received if the flow is lower than the flow specified in MinFlow. If an alarm is executed and the run is continued by pressing Continue, the sample loading will be stopped and the run will continue.	SampleFlow (0-50) Volume (0-1000) ReduceFlow No Yes MinFlow (OFF, 0.1-49.9) ml/min
PumpWashExplorer	PumpWash is used to change eluents in the inlet tubings, pump and mixer. THE PUMPS MUST BE PURGED PRIOR TO USING PUMPWASH. Flow is diverted to the waste position of the injection valve. Up to four inlets can be washed simultaneously; InletA1, InletA2, InletB1 or Inlet B2. One of the eight inlets of buffer valve A1 can be selected for InletA1. The system is set to Pause during PumpWash and the pressure limit is set to the value given in instruction Wash_alarm_pressure in <b>System:Settings</b> . After PumpWash the injection valve turns back to its previous position and the flow and pressure limits are set to their previous values.(Flow rate: 25 ml/min, Wash volume: 30 ml/inlet).	InletA1 OFF (A11-A18) InletA2 OFF ON InletB1 OFF ON InletB2 OFF ON
PumpWashPurifier PumpWashBasic	PumpWash instructions for ÄKTA <i>purifier</i> and ÄKTA <i>basic</i> , respectively.	-

---

SystemWash	<p>SystemWash is used to change eluents in the system, including inlet tubings, pumps, mixer and flow cells. THE PUMPS MUST BE PURGED PRIOR TO USING SYSTEMWASH. Flow is diverted to the waste at the outlet valve. The ColumnPosition is set to Position1Bypass during SystemWash. One inlet can be selected, A11-A18. When running BufferPrep methods the SystemWash instruction will deliver buffer with the selected composition (Recipe and BufferPrep_pH). The system is set to Pause during SystemWash and the pressure limit is set to the value given in instruction Wash_alarm_pressure in <b>System:Settings</b>. After SystemWash the OutletValve and ColumnValves turn back to their previous positions and the flow and pressure limit are set to their previous values.(Flow rate: 25 ml/min, Wash volume: 75 ml)</p>	InletA1 (A11-A18)
MethodBase	Sets the method volume calculation from either GradientPump or SamplePump	Base GradientPump SamplePump

## A.1.2 Flowpath

<i>Instruction</i>	<i>Description</i>	<i>Parameters</i>
InjectionValve	Sets the Injection Valve positions. The Injection Valve is switched to position 1 with parameter "load", to position 2 with parameter "inject" and to position 3 with parameter "waste". The instruction gives an injection mark when the injection valve switches to the "inject" position.	Position Load Inject Waste
ColumnPosition	Selects the position for the column. ColumnPosition switches the two column valves at the same time. Position1Bypass corresponds to a tubing connected to valve port 1 and Position2 corresponds to a column connected to valve port 2, etc. The flow rate is reduced to 0 ml/min when the valves switch.	Position Position1Bypass, Position2-8)
OutletValve	Selects the position for the Outlet valve. Feed switches the outlet valve one step forward from its actual position. The OutletValve gives fraction marks.	Position (WasteF1 FracF2 F3-F8 Feed)
BufferValveA1	Selects the position for Buffer Valve A1. Position A11 corresponds to position 1 on the Buffer Valve A1, position A12 to position 2, etc. The flow rate is reduced to 0 ml/min when the valve is switched.	Position (A11- A18)
PumpAInlet	Sets the position of the pump A inlet. The instruction is not allowed in a method when running BufferPrep (gives an alarm in BufferPrep mode).	Position A1 A2
PumpBInlet	Sets the position of the pump B inlet. The instruction is not allowed in a method when running BufferPrep (gives an alarm in BufferPrep mode).	Position B1 B2
SampleValve	Selects the position for the Sample-Valve.	Position (S1-S8)

---

FlowDirection	Sets the flow direction for the column. The instruction is useful for example when cleaning columns. The instruction switches the Flow Direction Valve to position 2 with parameter UpFlow and to position 1 with parameter DownFlow. The flow rate is reduced to 0 ml/min when the valve switches.	Direction Downflow Upflow
InjectionMark	The instruction gives an injection mark. The instruction is useful when the sample is introduced onto the column with the gradient pump.	-

### A.1.3 Frac

<i>Instruction</i>	<i>Description</i>	<i>Parameters</i>
Fractionation	<p>Starts fraction collection if the fraction size specified by the FracSize parameter is &gt;0. Tube change is delayed with respect to the set delay volume. FractionCollector is stopped with the instruction FractionationStop.</p> <p>Parameters:</p> <p>Tube type: The tube type to fractionate in</p> <p>FracSize: The size of the fractions</p> <p>Start at: Where to start the fractionation, if a specific position is desired use Tube number</p> <p>Base: The fractionation can be performed in time or volume base.</p>	<p>TubeType</p> <p>FracSize (0.000 - 99999.999)</p> <p>base</p> <p>StartAt (First tube</p> <p>Next tube</p> <p>Next line</p> <p>Next group</p> <p>Tube number)</p> <p>Base (Volume</p> <p>Time)</p>
FractionationStop	<p>Stops fraction collecting after the set delay volume has been collected. The flow is diverted to the waste of the outlet valve.</p>	-
FeedTube	<p>During Fractionation or Peak_Fractionation, FeedTube moves the tube rack forward one tube after the set delay volume has been collected. A fraction mark is given.</p>	-

Peak_Fractionation	<p>Peak_Fractionation enables only peaks to be collected. The UV monitor is used to determine when there is a peak to collect. PeakSize determines max volume to be collected in each tube. To collect the peak in one tube set PeakSize&gt;peak volume. If Peak_FracParameters are not set in the method, default values in system settings will be used. Tube change is delayed with respect to the delay volume. To stop Peak-Fractionation use the instruction Peak_FracStop.</p> <p>Parameters:</p> <p>Tube type: The tube type to fractionate in</p> <p>PeakSize: The size of the peak fractions</p> <p>Start at: Where to start the fractionation, if a specific position is desired, use Tube number</p> <p>Base: The peak fractionation can be performed in time or volume base.</p>	<p>TubeType</p> <p>FracSize (0.000-99999.999)</p> <p>base</p> <p>StartAt (First tube</p> <p>Next tube</p> <p>Next line</p> <p>Next group</p> <p>Tube number)</p> <p>Base (Volume</p> <p>Time)</p>
Peak_FracStop	Stops the peak fraction collection after the set delay volume has been collected.	-
Peak_Frac- Parameters	<p>UV1 Parameter settings for Peak_Fractionation. To obtain suitable values, perform a test run and measure the slope at the beginning and end of the smallest and flattest peak you want to collect, using</p> <p><b>Operations: Differentiate</b> in Evaluation. Time (min) as base must be used. Minwidth prevents collection of noise spikes. Enter a value of about half the width of the narrowest peak to collect.</p>	<p>StartSlope (0.01-10000 mAU/min)</p> <p>EndSlope (0.01-10000 mAU/min)</p> <p>MinWidth (0.01-1500 min)</p>
Reset_Frac_Position	Resets the position of the fraction collector for small and large tubes, and for start at next tube.	

---

OutletFractions	Enables fraction collection in subsequent outlets from the Outlet Valve. Fractionation starts at the selected outlet. If the number of programmed fractions exceeds the number of outlets, the fractionation will resume in position F3. After completing the last fraction the position selected before executing OutletFractions instruction will be executed.	How many? (0-7) Volume (5 - 100000 ml) Start at (F2-F8)
AccumulatorWash *	The accumulator makes 10 strokes, with 1 ml in each. The flow must be > 0 ml/min.	OFF ON
* When using the <b>AccumulatorWash</b> instruction for ÄKTAdesign systems, use manual control to begin flow and turn the Outlet valve to the frac position.		

## A.1.4 Alarms & Monitors

<i>Instruction</i>	<i>Description</i>	<i>Parameters</i>
Wavelength	Sets the wavelength and number of wavelengths to monitor. The range is 190-700 nm for UV1, UV2 and UV3. 0=OFF for UV2 and UV3. Wavelengths <190 nm are automatically set to 0=off.	UV1 (190-700 nm) UV2, 0=OFF (0-700 nm) UV3, 0=OFF (0-700 nm)
AutoZeroUV	Sets the relative AU to zero.	-
AveragingTimeUV	Filters the noise in the UV-signal. Averaging time is the time interval used for calculating the moving average of the absorbance signal. A long averaging time will smooth out noise efficiently, but will also distort the peaks. Choose an averaging time of the greatest value that can be set which is less than $(1/11) \cdot 60(\text{s/min}) \cdot \text{typical peak width (ml)} / \text{flow rate (ml/min)}$ . The averaging time should not be less than 1.28s when more than one wavelength is monitored. A column-specific averaging time is automatically set when a column and template are selected.	AvTimeUV 0.01 0.02 0.04 0.08 0.16 0.32 0.64 1.28 2.56 5.12
Alarm_UV1 Alarm_Cond Alarm_pH Alarm_Pressure Alarm_SamplePressure	Sets the alarm limits for the signal. An alarm will set the system in Pause.	Mode Disabled Enabled HighAlarm LowAlarm
SetCondScale 0%	Sets the 0% scale of the relative conductivity signal. The relative conductivity is displayed as a percentage of bufferB in run data (Cond%) and as a curve (Cond%).	CondValue (0-999.9 mS/cm)

SetCondScale 100%	Sets the 100% scale of the relative conductivity signal. The relative conductivity is displayed as a percentage of bufferB in run data (Cond%) and as a curve (Cond%).	CondValue (0-999.9 mS/cm)
WatchPar_UV1 WatchPar_cond WatchPar_Conc WatchPar_pH WatchPar_Pressure WatchPar_SamplePressure WatchPar_SampleFlow	The Delta_Peak setting affects only the Less_than_or_valley and Peak_max_conditions when using the Watch instruction (see UNICORN User Manual chapter 5.8.8). As a general guideline, set the value to 2-3 times the noise level and 5-10% of the smallest expected peak height. Delta_Base setting affects only the Stable_baseline condition. For this condition to be activated, the signal may not vary by more than the Delta_Base value above or below beyond the time interval specified in the Stable_baseline condition in the watch instruction.	Delta_Peak Delta_Base
UVlampOFF	Sets the UV lamp OFF. This command only influences the specific run.	-

### A.1.5 Watch

<i>Instruction</i>	<i>Description</i>	<i>Parameters</i>
Hold_Until	Puts the method on Hold until a specific condition (signal, test, or value) of the instruction is met, or the time out limit for the instruction is reached.	Signal (UV Cond pH Pressure) Test* (see below) Value TimeOut (0.0-999.9, INFINITE)

Watch_UV1 Watch_Conc Watch_pH Watch_Pressure Watch_SamplePressure Watch_SampleFlow	Sets a watch on the signal (UV1 for UV), to perform a set action if a given test condition is fulfilled. The watch is cancelled automatically when the condition has been fulfilled once.	Test * (see below) Value Action * (see below) Value
Watch_Off	Cancels a watch on a specified monitor.	Signal Pressure Conc UV pH Cond
*Test Greater_than Less_than Slope_greater_thanLess s_than_or_ valley Peak_max Stable_baseline	*Action Block names Ready Pause Hold End_Method End_Block Continue	

### A.1.6 Other

<i>Instruction</i>	<i>Description</i>	<i>Parameters</i>
Base	Defines the base for a block for calculating breakpoints. Each block must have a defined base.	Column (Any/ column name) Base Time Volume (ml) CV SameAsMain
Block	Calls a block unconditionally.	Block name
Continue	Resumes execution of a paused or held method. This instruction has the same effect as clicking on the Cont. button in System control.	-
End_Method	Terminates method execution, equivalent to clicking on the End button in System control.	-

Evaluate	Calls an evaluation procedure. The procedure must be stored together with the current method.	Procedure name
Hold	Places the system in Hold state. This instruction has the same effect as clicking on the Hold button in System control.	-
Loop	Runs the instructions between Loop and Loop_end for the specified number of times.	No of loops (1 - 9999)
Loop_end	Specifies the end of the instructions contained within a loop.	-
Message	Generates a user-defined message which is recorded in the log book and may be displayed on the screen.	"Message" Mode: Screen / Noscreen
New_chromatogram	A new chromatogram is created within the result file and all data collected after this instruction is stored within the newly created chromatogram until another New_chromatogram instruction is issued.	Name
Pause	Places the system in the Pause state for the specified length of time.	Time (-1 (infinite) - 9999.9 in minutes)
Ready	Indicates that the next step in a MethodQueue may start.	-
Set_Mark	Inserts a specified text into the chromatogram.	-
End_Block	Terminates a block and returns control to the point from which the block was called.	-
;	Inserts a comment in the method below the marked instruction.	Comment

---

## A.2 Manual control

### A.2.1 Pump

This group contains the same instructions as the Pump group in method instructions (Section A.1.1).

### A.2.2 Flowpath

This group contains the same instructions as the Flowpath group in method instructions (Section A.1.2).

### A.2.3 Frac

This group contains the same instructions as the Flowpath group in method instructions (section A.1.3), with the following instructions replacing the Frac instructions:

<i>Instruction</i>	<i>Description</i>	<i>Parameters</i>
Man_Fractionation	Starts fraction collection if the fraction size specified by the FracSize parameter is >0. Tube change is delayed with respect to the set delay volume. FractionCollector is stopped with the instruction FractionationStop. Parameter(s): Rack: The rack used. Frac_order: The fractionation order can be selected as Serpentine-row, Row-by-row, Serpentine-Column or Column-by-column. Tube type: The tube type to fractionate in. FracSize: The size of the fractions. Start at: Where to start the fractionation, if a specific position is desired, use Tube number. Base: The fractionation can be performed in time or volume base.	Rack Frac_order (Serpentine-row Row-by-row Serpentine-column Column-by-column Tube type FracSize (0.000 - 99999.999) base StartAt (First tube Next tube Next line Next group Tube number) Base (Volume Time)

Man_Peak_Fractionation	<p>Peak_Fractionation enables only peaks to be collected. The UV monitor is used to determine when there is a peak to collect. Peak size determines max volume to be collected in each tube. To collect the peak in one tube set PeakSize&gt;peak volume. If Peak_FracParameters are not set in the method, default values in system settings will be used. Tube change is delayed with respect to the delay volume. To stop Peak-Fractionation use the instruction Peak_FracStop.</p> <p>Parameters:</p> <p>Rack: The rack used.</p> <p>Frac_order: The fractionation order can be selected as Serpentine-row, Row-by-row, Serpentine-column or Column-by-column.</p> <p>Tube type: The tube type to fractionate in.</p> <p>PeakSize: The size of the peak fractions.</p> <p>Start at: Where to start the fractionation, if a specific position is desired use Tube number.</p> <p>Base: The peak fractionation can be performed in time or volume base.</p>	<p>Rack</p> <p>Frac_order</p> <p>(Serpentine-row</p> <p>Row-by-row</p> <p>Serpentine-column</p> <p>Column-by-column</p> <p>Tube type</p> <p>PeakSize</p> <p>(0.000 - 99999.999) base</p> <p>StartAt</p> <p>(First tube</p> <p>Next tube</p> <p>Next line</p> <p>Next group</p> <p>Tube number)</p> <p>Base</p> <p>(Volume</p> <p>Time)</p>
Ignore_LastTube	Allows for fractionation after the last tube position has been reached.	-

## A.2.4 Alarms & Monitors

This group contains the same instructions as the Alarms&Mon group in method instructions (Section A.1.4).

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## A.2.5 Other

<i>Instruction</i>	<i>Description</i>	<i>Parameters</i>
Block	Calls a block. Only available when a method is running.	Block Block name
Next_breakpoint	Jumps to the next breakpoint in the current method (only relevant when a method is running).	-
BufferPrepRecipe	Selects a recipe for running BufferPrep manually. The recipe must be selected as the first instruction for the manual run.	Recipe Name
Set_Mark	This inserts a specified text into the chromatogram.	-
End_Timer	This is used to set the time out limit, in time or volume, for the End instruction.	Accumulated time or volume 0-9999 min or 0-9999 l or disabled
Record_on	Begins recording a run that has been started manually. A result file will be generated.	-

## A.3 System settings instructions

### A.3.1 Alarms

<i>Instruction</i>	<i>Description</i>	<i>Parameters</i>
Alarm_UV1 Alarm_Cond Alarm_pressure Alarm_pH	Sets the alarm limits for the signal. An alarm will set the system in Pause.	Mode Disabled Enabled HighAlarm LowAlarm

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Wash_Alarm _pressure	Sets the alarm limits for pressure when instruction PumpWash or SystemWash are used. This pressure limit only applies when these instructions are executed.	Mode Disabled Enabled High Alarm 0-10 MPa Low Alarm 0-5 MPa
Alarm_SamplePressure	Sets the alarm limits for sample pressure. An alarm: SamplePressureHighAlarm/ SamplePressureLowAlarm will set the system in Pause.	Mode Disabled Enabled High Alarm 0-1.0 MPa Low Alarm 0-1.0 MPa

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## A.3.2 Specials

<i>Instruction</i>	<i>Description</i>	<i>Parameters</i>
FracParameters	Parameter settings for fraction collector. Delay volume is defined as the volume from UV flow cell to the outlet of the fraction collector. Collection of flow during tube change can be handled in different ways. Accumulator: The flow will be collected in the accumulator during tube change and then be delivered in the next tube. DropSync: Tube change synchronised to drop release. Should only be used at flow rates generating drops (below 5 ml/min if capillary 0.5 mm id is used, and below 3 ml/min for 0.25mm id tubing). Tube: No synchronisation of collection. Note! The delay volume must be changed when the parameter TubeChange is altered to or from Accumulator - see the manual for the correct delay volume.	DelayVol (0-10 ml) TubeChange (Accumulator DropSync Tube)
Peak_FracParameters	UV1 parameter settings for Peak_Fractionation. To obtain suitable values, perform a test run and measure the slope at the beginning and end of the smallest and flattest peak you want to collect, using Operations: Differentiate in Evaluation. Time (min) as base must be used. Minwidth prevents collection of noise spikes. Enter a value of about half the width of the narrowest peak to collect.	StartSlope (0.01-10000 mAu/min) EndSlope (0.01-10000 mAU/min) MinWidth (0.01-1500 min)
Keyboard	Sets the mode of the user interface of the instruments.	Mode Open KeyLocked KeyAndDial-Locked

### A.3.3 Monitors

<i>Instruction</i>	<i>Description</i>	<i>Parameters</i>
Wavelength	Sets the wavelength and number of wavelengths to monitor. The range is 190-700 nm for UV1, UV2 and UV3. 0=OFF for UV2 and UV3. Wavelengths <190 nm are automatically set to 0=off.	UV1 (190-700 nm) UV2, 0=OFF (0-700 nm) UV3, 0=OFF (0-700 nm)
AveragingTimeUV	Filters the noise in the UV-signal. Averaging time is the time interval used for calculating the moving average of the absorbance signal. A long averaging time will smooth out noise efficiently, but will also distort the peaks. Choose an averaging time of the greatest value that can be set which is less than $(1/11) * 60(\text{s/min}) * \text{typical peak width (ml)} / \text{flow rate (ml/min)}$ . The averaging time should not be less than 1.28s when more than one wavelength is monitored. A column-specific averaging time is set automatically when a column and a template are selected.	AvTimeUV 0.01 0.02 0.04 0.08 0.16 0.32 0.64 1.28 2.56 5.12
SetCondScale 0%	Sets the 0% scale of the relative conductivity signal. The relative conductivity is displayed as a percentage of bufferB in run data (Cond%) and as a curve (Cond%).	CondValue (0-999.9 mS/cm)
SetCondScale 100%	Sets the 100% scale of the relative conductivity signal. The relative conductivity is displayed as a percentage of bufferB in run data (Cond%) and as a curve (Cond%).	CondValue (0-999.9 mS/cm)

CondTempComp	Relates conductivity to concentration and/or compares conductivity values. The compensation consists of a compensation factor together with a reference temperature (CondRefTemp). All conductivity values will then automatically be converted to the set reference temperature. The factor is expressed in percentage increase of conductivity per °C increase in temperature. If the CompFactor is unknown, a general approximate value of 2% can be set for many common salt buffers. 0% = off.	CompFactor (0-9.9%)
CondRefTemp	Selects the reference temperature to which the measured conductivity values will be converted. CondRefTemp is active when a factor of CondTempComp is selected.	RefTemp (0-99.9°C)
pHTempComp	Sets the temperature compensation on or off. For more accurate measurements during temperature changes, the pH measurement can be temperature compensated. When using pHTempComp it is important that the temperature of the pH electrode is the same as that of the conductivity flow cell since that is where the temperature is measured.	pHTempComp ON OFF
WatchPar_UV1 WatchPar_Conc WatchPar_pH WatchPar_Pressure WatchPar_Sample- Pressure WatchPar_Sample- Flow	The Delta_Peak setting affects only the Less_than_or_valley and Peak_max_conditions when using the Watch instruction (see UNICORN User Manual). As a general guideline, set the value to 2-3 times the noise level and 5-10% of the smallest expected peak height. Delta_Base setting affects only the Stable_baseline condition. For this condition to be activated, the signal may not vary by more than the Delta_Base value above or below the time interval specified in the Stable_baseline condition in the watch instruction.	Delta_Peak Delta_Base

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### A.3.4 Curves

<i>Instruction</i>	<i>Description</i>	<i>Parameters</i>
UV1 UV2 UV3 Cond Cond% Conc pH Pressure Flow Temp Sample_Pressure Sample_Flow	Set the specific signal curve on/off for storing in the Result file. The time between samples determines the frequency with which curve data is recorded	Store ON / OFF Time between samples

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## **B Technical Specifications**

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### **B.1 System Requirements**

#### **B.1.1 Hardware requirements**

- Pentium II/333 MHz or later (minimum Pentium/90 MHz)
- 64 Mb RAM (minimum 32 Mb) for one system  
128 Mb RAM (minimum 64 Mb) for two or more systems
- 1 Gb of available hard disk space (150 Mb to run UNICORN), NTFS file system
- Colour monitor, 1024x768 pixels (minimum 800x600\*), small fonts, 64k colours
- 1 ISA or 1 PCI slot per connected system
- CD-ROM drive
- 1.44 Mb (3.5") diskette drive
- Mouse

\* option when industrially hardened screens are required

#### **B.1.2 Software requirements**

Microsoft Windows NT Workstation 4.0 (with Service Pack 5 or later).

#### **B.1.3 Network requirements**

These are the recommended network requirements for running UNICORN in a network installation:

- Supported Network cards:  
3COM Etherlink III  
Compaq Netelligent 10/100 TX Embedded UTP Controller  
Compaq Integrated NetFlex-3 Controller  
AMD PCNET PCI Ethernet Adapter (Integrated)
- Novell NetWare version 4.50.189 or later or Microsoft Windows NT Server 4.0. The UNICORN software works on earlier versions as well even though some versions of the Novell Netware driver have known problems.
- A valid network connection

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## **B.2 Control Capacity**

### **B.2.1 Stand-alone installations**

Simultaneous control of up to four chromatography systems. Each module is separately configured in a system strategy supplied by Amersham Biosciences.

### **B.2.2 Network installations**

A few basic facts about a network installation:

- Chromatography systems must be locally linked to a workstation and then the workstation is linked to the network, i.e. the chromatography systems are not directly linked up to the network.
- Each local workstation can be connected to four separate chromatography systems.
- A network can support up to 90 chromatography systems, which are connected locally to the workstations in the network.
- A workstation can locally or remotely actively control up to four chromatography systems. This is achieved using the four possible System control windows in UNICORN available on each workstation.
- Each chromatography system may be controlled by only one active System Control window, and be viewed by seven different other System control windows in UNICORN.

# C Installation of the PCI expansion card

*Information for service personnel:* In most cases, UNICORN is delivered pre-installed by qualified service personnel. The installation procedure described below should not be attempted by unqualified personnel.

The PCI expansion card replaces the ISA cards for the CU-900 control board supplied in earlier computer models. The upgrade is needed only for stand-alone installations (i.e. systems not connected to a network) and for local computers in network installations.

**Note:** This expansion card cannot be used with other CU units.

**Note:** Pentium 2 computers will require a BIOS update, available from: [www.compaq.com/support/files/desktops/us/index.html](http://www.compaq.com/support/files/desktops/us/index.html).

- 1. Turn off the power to the PC and unplug the main power cables.
- 2. Open the PC cover (see manufacturer’s instructions).
- 3. Locate an empty PCI expansion slot.

**Caution!** Electrostatic discharges can permanently damage electronic components on the card. Discharge any electricity from your body by touching an earthed metal surface. (e.g., a water tap or radiator) before handling the card.

- 4. Remove the expansion card from the anti-static bag. Handle the card by its edges, and, as far as possible, avoid touching the electronic components.

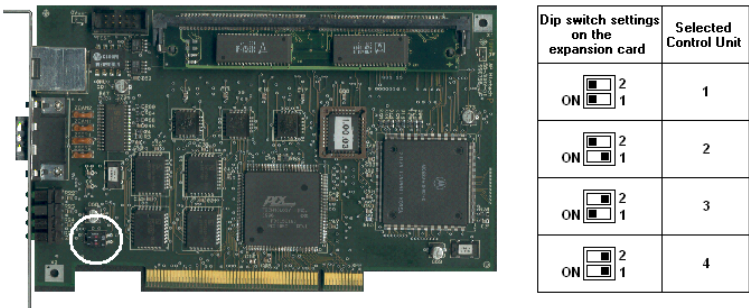


Figure C-1. The PCI expansion card and dipswitch settings for the CU.

- 
5. Set the control unit (CU) setting by using a dip switch. To check the CU setting, compare the expansion card with the settings shown in Fig. C-1. Record the number of the set CU. During installation of UNICORN software, the Setup program will request these card settings. Enter the correct value for the selected CU so that UNICORN can find the systems.
  6. Close the cover of the PC.
  7. Connect the expansion card to the liquid handling module CU connector using the communication cable provided. Additional liquid handling modules, up to a total of four, may be connected.

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